WEIGHT CURVES OF TUBERCULOUS GUINEA-PIGS

STUDIES ON THE BIOCHEMISTRY AND CHEMOTHERAPY OF TUBERCULOSIS. XX

LYDIA M. DEWITT

From the Otho S. A. Sprague Memorial Institute and Department of Pathology, University of Chicago

In most of the experimental work on the chemotherapy of tuberculosis, the apparently favorable results of treatment are based on the following: Increased duration of life, favorable influence on weight, and diminished distribution and progress of the disease. If it were possible in this disease in guinea-pigs to attain the ideal of all chemotherapy—complete sterilization of the animal with a single dose, it would be unnecessary to judge our success in this way. But so far no drug has been found which will completely destroy all the tubercle bacilli in a guinea-pig without also destroying the animal.

Much stress has been laid on prolongation of life as an effect of treatment. In 1917, however, it was shown by Paul Lewis [1] that the duration of life in any series of tuberculous animals is too variable to be used as an indication of therapeutic activity, unless the number of animals used is very large and the individual variations are completely accounted for. He states that with guinea-pigs, he has yet to conduct an experiment in which the last animal to die did not live at least twice as long as the first to die, and often the difference is much greater than this. No one who has worked with tuberculous guinea-pigs or with other animals having experimental tuberculosis has failed to note similar uncertainties and differences in the length of life of the animals in any given experiment, even though the animals had all received the same dose of the same strain of tubercle bacilli, the same treatment throughout and had lived under the same conditions. It is not always possible to account for these variations. Tables 1 and 2 give some of the variations in the duration of the disease in comparison with variations in weights.

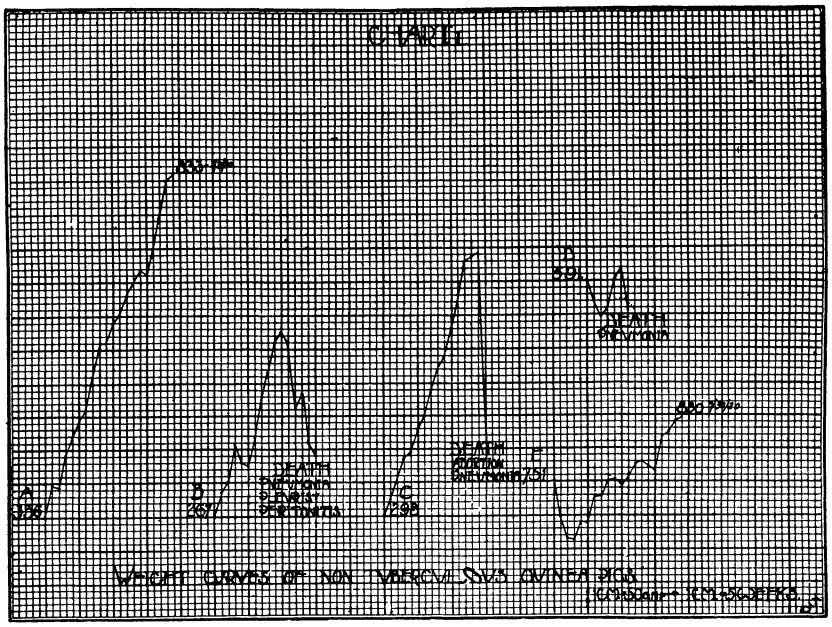
It is the purpose of this paper to endeavor to ascertain whether the weight curves of guinea-pigs inoculated with tuberculosis are any

[1] Fourteenth Report of the Henry Phipps Institute, 1918; Am. J. Med. Sc., 1917. 153. p. 625.

more uniform than the duration of life and, if not, whether the variations can be more satisfactorily explained.

For a number of years, it has been my custom to have all animals weighed once a week as a guide in treatment. Some of the earlier weights I have discarded in this paper and have used chiefly those sets the weighing of which I have personally supervised.

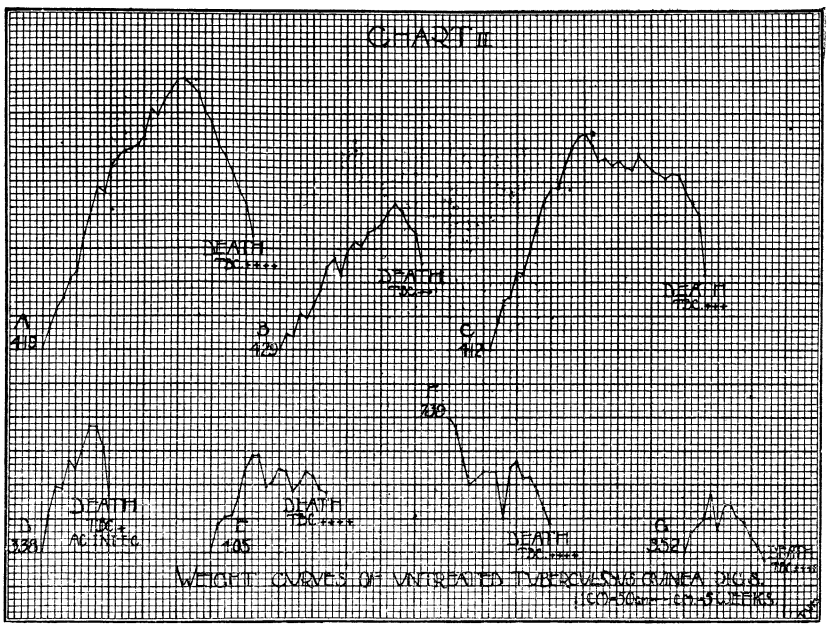
In order to have a basis of comparison, I have, during the last 22 weeks, been weighing once a week a number of nontuberculous and of supposedly normal guinea-pigs, the initial weight of which was



approximately the same as that of my inoculated animals. Chart 1 represents the type of weight curves of these animals with the cause of death in those cases in which death has occurred.

The curves in chart 1, as well as in the other charts, were made thus: The base line represents the weight of the animal at the time the experiment began—the initial weight line; this line is divided by vertical lines; one of the horizontal parallel spaces represents 10 gm. and one of the parallel vertical spaces represents one week. Nine supposedly normal guinea-pigs of different weights were used in this

experiment and were weighed once a week at the same time of the day, and, as far as possible, under the same conditions. Five of the animals died during the 22 weeks of this part of the experiment. Two died from infectious abortion with pneumonia, the weight curve (C, chart 1), showing a gradual rise as is typical of the normal growing animal and a sharp, sudden drop as seems typical of acute infections. Two, having a weight curve of the type of D, chart 1, died of an acute infection and one, having the curve B, chart 1, died from peritonitis and pneumonia. Of the 4 still living, 3 have a weight



curve typified by A chart 1, which seems to be the type of curve shown by normal, healthy, growing animals of medium initial weight. One of the heavy guinea-pigs, however, lost weight for about 3 weeks and then gradually gained and is still gaining (chart 1 E).

Chart 2 reproduces 7 of the most typical weight curves found in 37 tuberculous but untreated guinea-pigs. Table 1 itemizes the history of the individual animals used for chart 2, in which it may be noted that A, B, C and D are similar in contour, differing in height, in form of the summit, or in the width of the elevation according to

the rapidity of the ascent or descent and the length of life. An acute infection sharpens the summit and narrows the entire elevation. One of these four types is found 29 times among the 37 animals in table 1.

TABLE 1

TABLE OF WEIGHTS AND DURATION OF LIFE OF UNTREATED TUBERCULOUS GUINEA-PIGS

No. of Set	Duration of Disease, Days	Sex	Initial Weight, Gm.	Greatest Weight, Gm.	Terminal Weight, Gm.	Duration of Ascent, Weeks	Duration of Descent, Weeks	Type of Curve	Degree of Tuberculosis	Notes
1	129	F.	405	548	484	7	10	2 E	++++	
2	123	M.	386	569	408	10	6	2 A	++++	
1	131	M.	375	509	357	11	7	2 A	++++	
1	145	M.	437	589	465	10	4	2 C	++++	
1	158	M.	429	640	508	17	4	2 B	++++	
1	223	M.	419	815	589	20	11	2 A	++++	
1	78	M.	338	534	432	8	2	2 D	+	Acute infection
1	143	M.	336	605	412	8	5	2 C	++++	
1	134	M.	481	635	394	6	10	2 C	++	
1	113	M.	345	605	531	12	3	2 D	+++	
1	98	M.	360	520	436	8	6	2 B	++++	
1	246	M.	412	730	522	15	10	2 C	++++	
1	158	M.	306	568	340	15	6	2 A	++++	
1	167	M.	455	671	440	14	8	2 A	++++	
1	130	M.	430	686	457	11	5	2 D	++++	
1	116	M.	454	612	495	8	7	2 D	++++	
1	97	F.	388	533	368	5	7	1 B	+++	
1	103	M.	410	497	422	5	6	2 G	++++	
2	167	M.	352	599	432	15	9	2 A	++++	
2	169	M.	382	563	417	15	9	2 B	++++	
2	164	M.	440	674	608	15	3	2 C	++++	
2	171	M.	484	730	495	10	14	2 C	++++	
2	113	M.	375	467	326	7	9	3 J	++++	Acute infection
2	101	M.	287	441	335	9	6	2 D	++++	
3	74	F.	266	427	321	8	3	2 D	++++	
3	74	M.	264	384	292	8	3	2 D	+++	Acute infection
3	148	M.	215	401	384	12	9	2 E	+++	Killed
3	112	M.	177	344	274	14	2	2 C	++++	
3	109	M.	259	391	313	10	5	2 A	++++	
4	27	F.	590	680	660	2	1	2 D	++	Acute infection
4	60	F.	240	290	205	4	2	3 B	++++	
4	104	M.	295	425	255	10	3	3 C	++++	
4	105	M.	580	685	610	7	3	2 E	++++	
4	105	F.	730	730	500	0	15	2 F	++++	
5	82	M.	352	438	341	4	6	2 G	+++	
5	76	M.	352	395	309	5	5	2 G	++++	
5	69	M.	378	410	310	2	6	3 A	++++	

Set 1 of table 1 consists of 18 guinea-pigs, varying in weight between 350 and 450 gm., which were inoculated at the same time with the same amount of the same strain of human tubercle bacilli. Six received the ordinary laboratory feeding of carrots, oats and hay. Twelve received lettuce and specially prepared graham crackers and hay. Hence the animals of set 1 differed only in their food. The weight curves of 16 of the 18 can be classed under the 4 types which I regard as typical for tuberculosis in guinea-pigs. One had the weight curve E, chart 2, which differs from C only in the fact that

death occurred very near the summit of the elevation and that the ascent was not so high. One had the curve G which differs from B in being lower and in having a double apex. Set 2, table 1, is composed of the 6 untreated controls of our cresol experiments. They were inoculated with the same strain of human tubercle bacilli as was set 1, but received four times as large a dose. Five of these had curves of one of the four main types, but one, which developed an acute peritonitis in addition to the tuberculosis, showed a curve similar to J, chart 3, which is formed much like the typical curve, but is lower and descends below the base line.

Set 3 consisted of 5 guinea-pigs inoculated with the washings from garnets used as controls in a bactericidal experiment. The garnets were covered with an unkonwn quantity of tubercle bacillus suspension, then exposed to salt solution, washed and shaken in salt solution. Hence the dose used in the inoculation was unknown. However, of the 5 animals whose curves were taken, 4 had the typical curves A, C and D, while one had curve E, in that it was killed near the apex of the curve. Set 4 was inoculated with an unusually large dose of a very virulent strain. As a result, the duration of the disease was relatively short and the curves less typical than in the other sets. Set 5 was also a bactericidal control. Only three curves were used, since most of the animals are still living. Curve G reproduces the curve in two of these. The sudden drop shown in this curve was due to the fact that we had had no carrots for 4 days, so that during this time they had lived on oats and hay. The same explanation applies to curve A, chart 3, which more nearly resembles the curve of the third animal. In studying table 1 and chart 2 in the effort to determine the causes of the typical curves and of any modifications from the types, we note that most of the animals were young adults, still growing but past some of the weaknesses which belong to the young. Extreme age, as indicated by 2F and 1E, tends to modify the curve considerably. The natural tendency of the weight curve in the normal growing animal is constantly upward, as shown in A, chart 1, and since the inoculation with a small or moderate dose of a moderately virulent strain of human tubercle bacilli does not materially influence health at first, the curve follows its natural upward tendency until something in the way of diet or secondary infection or an overpowering of the animal with the effects of the tubercle bacillus impairs the health and the curve begins to descend. Larger initial doses or a more

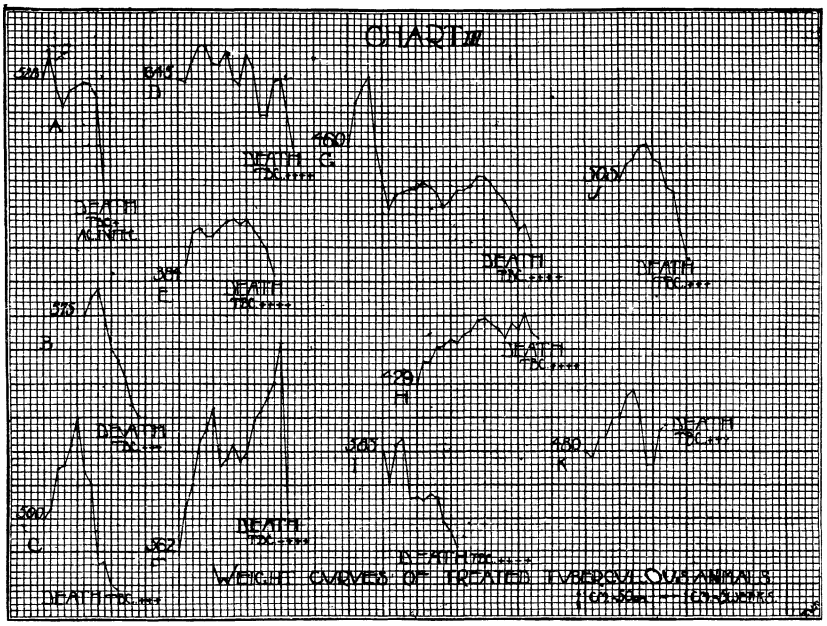
virulent strains of organisms tend to cause shorter ascending curves and thus a lower elevation. Secondary infections may cause sharp, sudden descent. Sex seems to have some effect on the shape of the curve, since of the 6 females in table 1, 4 had more or less atypical curves, while only 8 of the 30 males had curves varying in any way from the typical.

It is not always possible to explain variations. It is even difficult to explain the typical curve. The ascent is easy to explain since that is natural. But why the occasional sharp breaks in the ascent? Why the final slow or rapid descent? Why the sharp or rounded apex in some cases and the broad but jagged plateau in others. We have already seen how variations in the diet may cause decided drops in the weight and these or slight infections may explain the occasional jagged outline of the ascent. The final descent does not necessarily mean the generalization of the tuberculosis, since several animals that died or were killed at or near the apex of the curve showed all the internal organs involved in the tuberculous process. We must, therefore, assume that the descent is due to the overpowering of the animal by the toxic products of the bacteria. Whether the apex shall be broad or narrow probably depends on whether the resistance gives way suddenly or gradually—in other words, on the individual factor in the equation.

The question of the effect of treatment on the weight curve can be answered only so far as it concerns the 56 cases whose weight curves have been plotted. No generalization can be made from this number since it is self-evident that the effect of treatment on the weight curve as well as its effect on the condition of the animal, on the progress of the disease and on the length of life must vary with the treatment. Each drug used will probably affect the curve differently.

Chart 3 gives in graphic form some of the types of curves found in the 56 animals which had received six different modes of treatment. The curves were plotted as in charts 1 and 2. Table 2 gives in detail the individual animals of these various sets, with the treatment of each, the duration of life, the weights and the type of weight curve shown, the numeral referring to the chart and the letter to the curve. Many of the curves varied so little from those of the untreated animals that the type in chart 2 or 1 can be used in describing these treated animals. Chart 3 therefore reproduces in the main those types

that are quite different from those curves in the untreated animals, i. e., atypical curves. As may be seen from table 2, four of the 6 guinea-pigs treated with orthocresol had typical tuberculosis curves and only 2 the less typical curves F and G of chart 3. Metacresol and paracresol treatments, on the other hand, give only 2 characteristic curves from chart 2, and 4 curves from chart 3. Two of the guinea-pigs treated with thymol, which died of an acute infection and had little tuberculous involvement had weight curves similar to I and J of chart 3, three had typical weight curves and one, which is still living, has so far the typical



weight curve of a normal animal, as represented in A of chart 1. Of 6 animals treated with mercurophen, 3 had typical curves of untreated animals, while 2 had a low elevation shaped like the typical curve and 1 had a triple crested curve like E of chart 2. Of the 11 animals treated with the double salt of mercuric chlorid and methylene blue and the 8 treated with the same double salt combined with a vaccine of human tubercle bacilli, only 1 was typical, all the others showing some one of the abnormal types in chart 3. Five tuberculous guinea-pigs treated with the double salt of mercuric chlorid and methylene blue are still

TABLE 2

TABLE OF WEIGHTS AND DURATION OF LIFE OF TREATED TUBERCULOUS GUINEA-PIGS

Duration of Disease, Days	Sex	Initial Weight, Gm.	Greatest Weight, Gm.	Terminal Weight, Gm.	Duration of Ascent, Weeks	Duration of Descent, Weeks	Type of Curve	Degree of Tuberculosis	Treatment
113	F.	562	879	580	10	3	3 F	++++	Orthocresol
188	F.	460	562	314	13	12	3 G	++++	Orthocresol
113	M.	553	664	475	8	9	2 B	++++	Orthocresol
87	F.	427	613	401	9	4	2 D	++++	Orthocresol
142	F.	586	822	754	19	2	2 B	++++	Orthocresol
118	M.	324	480	387	8	9	2 A	++++	Orthocresol
98	M.	355	551	434	8	5	2 A	++ *	Metacresol
74	F.	575	654	436	7	4	3 J	++++	Metacresol
87	M.	362	535	425	9	4	2 B	++ *	Metacresol
107	M.	415	513	440	5	3	3 E	+++	Metacresol
80	F.	585	611	425	2	6	3 I	++++	Metacresol
73	M.	503	554	357	4	6	3 J	+++	Metacresol
170	M.	415	556	440	14	8	2 C	+++	Paracresol
120	M.	389	491	373	10	4	3 E	+++	Paracresol
134	M.	429	534	499	16	2	3 H	+++	Paracresol
162	M.	390	534	400	12	5	3 E	+++	Paracresol
134	M.	344	561	457	17	2	2 B	++ *	Paracresol
58	M.	455	556	402	3	5	3 J	+ *	Thymol
80	M.	483	554	386	6	6	3 I	++ *	Thymol
115	M.	396	547	419	10	6	2 C	++ *	Thymol
136	M.	390	553	394	8	12	2 B	++++	Thymol
147	M.	357	486	337	9	11	2 A	+++	Thymol
	M.	413	...	...	..	..	1 A	.....	Thymol
89	M.	420	490	442	6	4	2 E	++ *	Mercurophen
139	M.	368	650	617	16	3	2 C	++++	Mercurophen
71	M.	367	501	387	8	2	2 D	+ *	Mercurophen
107	M.	430	551	493	7	1	2 C	+ *	Mercurophen
97	M.	384	465	383	10	4	3 E	+++	Mercurophen
132	M.	428	549	397	12	6	3 E	++++	Mercurophen
120	F.	380	515	435	11	6	3 F	+++	$HgCl_2$ methylene blue
114	M.	510	665	505	12	5	2 B	++ *	$HgCl_2$ methylene blue
68	F.	565	610	390	1	6	3 B	+++	$HgCl_2$ methylene blue
87	F.	500	650	385	5	6	3 C	++++	$HgCl_2$ methylene blue
75	F.	575	615	425	3	7	3 B	+++	$HgCl_2$ methylene blue
68	F.	480	570	520	9	3	3 K	+++	$HgCl_2$ methylene blue
76	M.	480	515	335	5	5	3 B	+++	$HgCl_2$ methylene blue
56	F.	440	455	330	2	5	3 I	+++	$HgCl_2$ methylene blue
75	F.	585	710	510	4	4	3 G	+++	$HgCl_2$ methylene blue
62	M.	528	565	358	4	6	3 A	+ *	$HgCl_2$ methylene blue
73	M.	572	572	364	3	6	2 F	+ *	$HgCl_2$ methylene blue
142	M.	450	550	445	8	8	3 E	++++	Tuberculosis vaccine and $HgCl_2$ methylene blue
129	M.	695	765	545	6	10	3 F	++++	Tuberculosis vaccine and $HgCl_2$ methylene blue
156	F.	620	665	530	5	5	3 G	++	Tuberculosis vaccine and $HgCl_2$ methylene blue
143	M.	645	695	535	6	9	3 D	+++	Tuberculosis vaccine and $HgCl_2$ methylene blue
153	M.	690	725	650	7	6	3 A	+++	Tuberculosis vaccine and $HgCl_2$ methylene blue
85	F.	520	540	410	2	8	3 B	++++	Tuberculosis vaccine and $HgCl_2$ methylene blue
66	M.	685	710	555	3	4	3 B	—	Tuberculosis vaccine and $HgCl_2$ methylene blue
110	F.	445	525	495	12	3	2 E	++++	Tuberculosis vaccine and $HgCl_2$ methylene blue
60	F.	575	575	325	0	10	2 F	++++	Iodine in starch
100	M.	675	685	560	4	7	3 J	++++	Iodine in starch
78	M.	560	620	510	4	5	3 J	++++	Iodine in starch
77	F.	625	625	470	3	6	2 F	++++	Iodine in starch
90	M.	415	535	335	7	6	2 E	++++	Iodine in starch
95	M.	470	530	395	5	8	2 G	++++	Iodine in starch
138	M.	409	469	334	9	9	3 E	++++	Iodine in milk
117	M.	341	462	321	8	8	3 E	++++	Iodine in milk

* Acute infection.

living, but their weight curves up to the present time do not conform to the types that we have regarded as typical for untreated tuberculous animals. Two sets of animals have been treated with iodin. In the first set, the iodin was given in powdered starch and in these the curves were all more or less atypical. Of the second set, which have been fed iodin in milk powder, only two have died, showing the low but typically shaped curve represented in E of chart 3. Five of the guinea-pigs of the second set are still living and so far their curves correspond with the normal curve A in chart 1 or with the beginning of A or C of chart 2.

In order to determine how much of the effect on the weight curve in the treated animals was due to the treatment and how much to the treatment combined with the tuberculous infection, 3 normal guinea-pigs have received the same treatment as each set of pigs given in table 2 with the exception of the iodin in powdered starch and the tubercle bacillus vaccine combined with the double salt of mercuric chlorid and methylene blue. Of the 21 normal guinea-pigs thus treated whose weight curves have been thus charted, 4 have died of acute infections and have the curve of most acute infections. The rest are living after 3 and 5 months' treatment and their weight curves correspond with A of chart 1. In other words, if uninfected, they run, in spite of the treatment, the typical weight curve of the normal animal; that is to say, while these drugs modify considerably the weight curves of tuberculous animals, they have not, during the months of the experiment, changed the weight curves of normal, uninfected guinea-pigs. In the 37 untreated tuberculous animals, the height of the elevation, i. e., the difference between the initial weight and the maximum weight averaged 166 gm. and, if sets 4 and 5 of table 1 be omitted because of the larger dosage and greater virulence of the infectious organisms, the average height of the elevation is 195 gm. In the 55 treated tuberculous guinea-pigs, on the other hand, the average height of the elevation was only 100 gm. If, however, we wish to compare the average elevations of the tuberculous animals under the different methods of treatment, we find the average weight elevation of the 22 animals treated with the cresols and thymol is 139 gm.; the 6 tuberculous animals treated with mercurophen have an average weight elevation of 135 gm. The tuberculous animals treated with the double salt of mercuric chlorid and methylene blue have an average weight elevation of only 75 gm., while that of the 8 animals

treated with the same double salt and a vaccine of killed tubercle bacilli is 53 gm. Fifty-four grams is the average weight elevation of the 8 iodin-treated pigs.

If we compare the treated animals with the untreated with reference to the descent of the weight curve below the line of the initial weight, we find that only 11 of the 37 untreated animals had curves descending below the base line, while 37 of the 55 treated animals had such long descending curves. The average distance of the terminal weight below the initial weight in the untreated animals was 56 gm., while only 1 animal had a curve descending more than 100 gm. below the base line. The treated animals, however, showed an average distance below the base line of 95 gm., while 18 of the 37 had curves descending more than 100 gm. below the base line.

CONCLUSIONS

Normal guinea-pigs of approximately the same age and weight and living under the same conditions run a uniform weight curve. This curve is easily modified by changes in diet, by acute infections and other variations in the conditions of life.

Normal male guinea-pigs of approximately the same age and weight inoculated with the same dose of the same strain of tubercle bacilli and living under the same conditions, run a fairly uniform and typical weight curve. This weight curve may therefore be used in testing the effect of various methods of treatment and is a more reliable standard than the duration of life.

Most chemotherapy, so far as tested, even though the drugs and doses used are so nontoxic as not to interfere materially with the duration of life or with the weight curves of normal, uninfected guinea-pigs, tends to alter materially the type of weight curve. This alteration consists in the main in a diminution in height of the ascending curve and an increase in length of the descending curve.

It may be inferred that the more closely the weight curves of tuberculous animals treated by any method adhere to the normal weight curve, the more benefit we may hope for from the treatment.